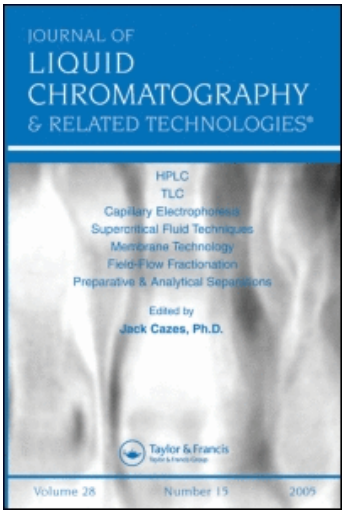

Journal of Liquid Chromatography & Related Technologies

Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

Simultaneous Separation of Inorganic Ions and Neutral Organics on a Sol-Gel-Coated Capillary using Combined Capillary Zone Electrophoresis and Capillary Electrochromatography

John R. Jezorek[a]; Rong-Fang Gu[a]; Tricia H. Dean[a]; Chantel L. Sabus[a]; Xin Sha[a]
[a] Department of Chemistry and Biochemistry, The University of North Carolina at Greensboro, Greensboro, North Carolina, USA

To cite this Article Jezorek, John R. , Gu, Rong-Fang , Dean, Tricia H. , Sabus, Chantel L. and Sha, Xin(2008) 'Simultaneous Separation of Inorganic Ions and Neutral Organics on a Sol-Gel-Coated Capillary using Combined Capillary Zone Electrophoresis and Capillary Electrochromatography', Journal of Liquid Chromatography & Related Technologies, 31: 19, 2942 — 2954

To link to this Article: DOI: 10.1080/10826070802424535

URL: http://dx.doi.org/10.1080/10826070802424535

Journal of Liquid Chromatography & Related Technologies®, 31: 2942–2954, 2008

ISSN: 1082-6076 print/1520-572X online
DOI: 10.1080/10826070802424535

Simultaneous Separation of Inorganic Ions and Neutral Organics on a Sol-Gel-Coated Capillary using Combined Capillary Zone Electrophoresis and Capillary Electrochromatography

John R. Jezorek, Rong-Fang Gu, Tricia H. Dean, Chantel L. Sabus, and Xin Sha

Department of Chemistry and Biochemistry, The University of North Carolina at Greensboro, Greensboro, North Carolina, USA

Abstract: Capillary zone electrophoresis was combined with capillary electrochromatography to effect the separation of ionic and neutral organic mixtures. Sol-gel technology was used to coat the capillary wall with a silane-based stationary phase. An n-octyl phase was used for cation-organic mixtures and an octadecyl phase, which incorporated a quaternary ammonium group to effect electroosmotic flow reversal, was employed for anion-organic mixtures. Tetraethoxy silane was used as a spacer to control the density of stationary phase interactive groups. Some data on the optimum ratio of analyte-interactive group to spacer silane in the sol-gel reaction mixture was obtained. Mobile phases were mixtures of typical capillary electrophoresis run buffer and reversed phase chromatography constituents. In all cases, the analytes were separated both within and between classes, the ions exiting the capillary as a group before the neutral organics. Investigation of mobile phase methanol cosolvent and pH effects found that electrophoretic and chromatographic behavior in the mixed mode system was what would be observed in a single mode separation, demonstrating the independence of the two modes. Separation of ions and neutrals in a red wine and multivitamin is demonstrated.

Correspondence: John R. Jezorek, Department of Chemistry and Biochemistry, The University of North Carolina at Greensboro, P.O. Box 26170, Greensboro, NC 27412-6170, USA. E-mail: jjezorek@uncg.edu

Keywords: Capillary electrochromatography, Capillary electrophoresis, Mixed class separation, Simultaneous ion-neutral separation, Sol-gel coating

INTRODUCTION

Chemical separation methods typically are designed to access only one analyte class, for example inorganic ions or neutral organic compounds. However, an ability to separate and determine analytes both within and between classes in the same run might afford both reduced analysis time and cost. For some time our laboratory has worked on modifying existing single class liquid chromatographic procedures to address the separation and detection of two or more analyte classes in one run. Both weak[1] and strong[2,3] ion exchange columns have been employed to simultaneously separate mixtures of inorganic ions and both polar and nonpolar neutral organic molecules. Another approach which our laboratory has used is that of ion interaction HPLC in which a standard reversed phase column is dynamically coated with a charged surfactant contained in the mobile phase, affording ion exchange or ion interaction properties to the system while retaining the reversed phase interactions. We have used this approach for both mixtures of metal cations and neutral organics,[4] as well as inorganic anions and neutral organics.[5] The above references also describe work by other laboratories, which have employed serial, mixed bed and mixed mode columns to separate different analyte classes.

A liquid phase separation method that has gained attention in recent years due to the promise of efficiency considerably greater than that afforded by conventional packed column liquid chromatography is that of capillary electrophoresis (CE). Historically applied to charged analytes in the capillary zone electrophoresis mode (CZE), CE can also be used to separate neutral species if a miscellar pseudo stationary phase is employed in the running buffer. This approach is called miscellar electrokinetic capillary chromatography (MEKC).[6] A variation on this theme is to permanently or dynamically coat the wall of the capillary with a true stationary phase, permitting capillary electrochromatography (CEC), wherein the electroosmotic flow (EOF) provides the mobile phase movement.[7] This CEC approach retains most of the high efficiency advantage of open tubular columns, unlike packed capillaries which degrade efficiency due to the particles and required end fitting frit. With silicon based wall coatings the electrophoretic ion separating character remains, opening up the possibility of marrying CZE to CEC and the simultaneous separation of both ionic and neutral species in one run. Li, Fries, and Malik have published a rather comprehensive article reviewing the use of CEC, as well as sol-gel technology to produce stationary phases.[8] The present report describes our efforts to use sol-gel technology to coat

the capillary wall and to separate mixtures of inorganic anions and neutral organics, as well as metal cations and neutral organic species.

EXPERIMENTAL

Instrumentation

Separations of anion–organic mixtures were done on a GTI/Spectrovision Capillary Electrophoresis System using a DA-30R High Voltage Power Supply. Metal ion-organic mixtures employed a Spectrophoresis 100 capillary electrophoresis system from Thermo Separation Products. Online detection used either a Linear Model 200 or Linear Model 525 variable wavelength detector. Detection for anion-organic mixtures was at a set wavelength of 250 nm, while cations were indirectly detected at about 220 nm and organics around 250–260 nm in cation-organic mixtures. The Model 525 detector was programmed to change to the higher wavelength once all the metal cations had been detected. Data acquisition employed either EZ Chrom software, version 6.7, or EZ Chrom Elite, version 2.61.

Materials

The fused silica column material was 360 μm outside diameter, either 75-or 25-μm inside diameter, and was obtained from Polymicro Technology or Supelco. The detection window was obtained by burning away the polyimide coating and cleaning the capillary with methanol to remove charred material. Generally a length of bare fused silica capillary was used to extend the length of shorter pieces of modified capillary. Connection of the two lengths was made with a universal capillary connector (Alltech). The detection window was usually in the unmodified portion. This arrangement allowed shorter lengths of modified capillary ($\approx$ 30 cm) to be tested but left the piece of capillary with the detection window ($\approx$ 45 cm) in place, minimizing breakage at the window. The end of the capillary was dipped into hexane for 10 s to soften the polyimide coating and effect a tight seal to the connector. Total column length to the detector window was about 48 cm. A 10 mL reaction vessel (Fisher) was used to prepare the sol-gel toward the end of the study.[9] This has an inlet that allows gas pressure to be applied to force the sol-gel mixture into the column.

The tetraethoxysilane (TEOS) was obtained from Lancaster Synthesis; the octyltriethoxysilane (C8-TEOS) was obtained from Lancaster Synthesis (95%) or from Aldrich (96%). The N-octadecyldimethyl

[3-trimethoxysilylpropyl] ammonium chloride (ODQAS) was purchased from Petrarch (Hülls). All analyte solutions and run buffers were made up in water purified with a Nanopure (II) system (Barnstead). Reagents were either HPLC or ACS reagent grade. All solvents (water and methanol), run buffers, and analyte solutions were filtered through 0.45μm nylon or polypropylene membrane filters. Analyte solutions were generally in the millimolar region. Buffer concentration in samples was typically about 10% that of the run buffer.

Operating Conditions

Running voltage was in the 20–25 kV range and was unchanged during a run. Injection was done electrokinetically, typically at 20 kV for 3 s. Run buffer and waste vials were filled with fresh solution at the start of each day and after 3–4 hours of running. Run buffer solutions were degassed with helium if bubbles were a problem. Columns were flushed with water and run buffer for 5 min, then equilibrated with run buffer for 30 min at the running voltage for the first run of the day, and flushed with run buffer for 2 min between runs. Electrodes and capillary inlet were rinsed with fresh run buffer after each injection to remove excess sample. Capillaries were flushed with water and run buffer at the end of the day and stored overnight filled with water or run buffer.

Column Preparation

The bare fused silica capillary to be coated was pretreated by being flushed with 0.1 M NaOH for 30–60 min and then water for 30–60 min. The remaining fluid was forced out with helium at about 12 psi for 15–30 min and the capillary was dried overnight under vacuum at 120°C. The sol-gel was prepared in a small beaker or the 10 mL reaction vessel by mixing the appropriate volumes of TEOS, ethanol, water, 0.1 M HC1, and either ODQAS (for anion–organic work) or C8-TEOS (for cation–organic work). The mixture was stirred for a given amount of time (e.g. 6 hr) at ambient conditions. If the sol-gel preparation was successful the mixture remained clear and colorless. The pretreated capillary was filled either with a syringe and allowed to stand for 10 min, or using helium pressure at 12 psi was introduced from the 10 mL reaction vessel and run through the column for 5 min, and allowed to stand for 5–10 min. The excess sol-gel was removed with helium pressure (12 psi) for 10–15 min. The capillary was sometimes examined under a microscope to be sure the channel was not blocked. Curing of the wall coated capillary was done overnight at 120°C under vacuum.

RESULTS AND DISCUSSION

In order to separate inorganic ions and neutral organic species in one run, differential interaction mechanisms for each class of analyte must be present. Therefore, in addition to the mass: charge electrophoretic separation mechanism (CZE), a stationary phase must be introduced for the neutral species (CEC). As mentioned above, the latter can be present in the run buffer/mobile phase in the form of micelles,[6] or as a conventional stationary phase packing or wall coating.[7] In order to retain the efficiency advantage of open tubular capillaries, this study employed a wall coated stationary phase put in place using sol-gel technology.[8]

As we wanted to investigate both anion-organic and cation-organic mixtures, we needed to employ two different capillaries with distinct stationary phases. In order to provide electrophoretic mobility toward the detector window it is necessary to use a polarity opposite that of the analyte ions for the downstream electrode. This means placing the cathode (negative) at the capillary end for cation separation, and the anode (positive) at the end for anions. However, in order to also have the EOF in the same direction as the analyte ions the capillary wall or stationary phase coating must have immobile charges opposite that of the analyte ions. For cations, residual silanol (SiOH) sites fulfill this requirement, as above pH 3 the silanol protons are ionized providing negatively charged sites. For anion separations, however, it is necessary to design a stationary phase that provides a permanent positive charge. For this we chose a quaternary-ammonium moiety to be incorporated into the stationary phase wall coating, as described below.[10] Of course, for neutral organic analytes the stationary phase must contain sufficient dispersion force sites to differentially interact with those molecules.

Column Preparation

As mentioned above, we chose the sol-gel method to apply the stationary phase to the capillary wall.[8] This approach utilized a silane material containing reactive groups for wall attachment, the interactive sites for organic analyte separation, and in the case of anion-organic mixtures, a permanent positive charge for EOF reversal. A solvent, ethanol, was used along with a stoichiometric amount of water to hydrolyze the alkoxy reactive groups, and a catalyst, hydrochloric acid, in our case.

The reaction involves, first, hydrolysis of the alkoxy groups, ethoxy in our case, polycondensation of the silane monomers, then deposition by reaction with capillary-wall sites. Finally, the wall coating is cured to drive off the trapped water and solvent and to complete the formation of the porous glass coating, the stationary phase.

The actual silanes used in this work were n-octyl-triethoxysilane (C8-TEOS) for cation–organic mixtures and N-octadecyldimethyl[3-trimethoxysilylpropyl] ammonium chloride (ODQAS) for anion–organic analyte mixtures.[8,9] In addition, tetraethoxy silane was employed as a spacer co-precursor to control the density of the stationary phase interactive sites. Relative amounts of all reaction constituents generally followed published procedures.[8,9]

The literature indicated that a spacer was advantageous for efficient separation of neutral analytes,[8] but that the optimum ratio of spacer precursor to that containing the analyte interacting moiety was in doubt.[11–13] We decided to do our own studies with C8-TEOS and TEOS. Our first effort involved ratios of C8-TEOS–to–TEOS of 0.4, 0.6, 0.8, and 1.2. Each column was prepared in triplicate using 420 μL of TEOS (2.02×10^{-3} mol), 172 μL of ethanol solvent, about 40 μL of 0.1 M HC1, and the appropriate number of moles of C8-TEOS. The amount of water used was $4 \times$ moles of TEOS $+ 3 \times$ moles of C8-TEOS. Phenol and nitrophenol were used as test analytes, and plots of capacity ratio and resolution for both reached a maximum value at a ratio about 0.8, with only slight increases beyond that, in general agreement with results from the Colon group.[12,13] We decided to repeat this study except using less acid catalyst (10 μL of 0.1 M HC1) and less water ($2 \times$ moles TEOS $+ 1.5$ moles C8-TEOS).[11] Ratios of C8-TEOS–to–TEOS from 0.1 to 0.8 were used, and the entire series was performed three times, totally from scratch each time. Again, phenol and nitrophenol were the test analytes. Capacity ratio and resolution were again used as markers. This time, however, k and R reached a maximum at about 0.2 and decreased after that. This can be seen in the sample chromatograms from one of the three ratio studies (Figure 1), where trace B is obviously superior to the others. These results are more in line with those reported by Constantin and Fritag,[11] who found a k maximum at a ratio of about 0.3.

Some preliminary investigations were also done on wall coating stability and on column preparation reproducibility. In the early stages of this work capillaries were filled with the sol-gel manually using a syringe. Reproducibility from one capillary to the next was very poor. While all capillaries were able to separate ions and neutral organics, differences in retention and efficiency were rather large.[9,12] Relative differences in k for naphthalene and phenanthrene, for example, were 5% and 14%, respectively, from one column to the next, prepared in the same way on two consecutive days. A more reproducible way of introducing the sol-gel into the capillary and of removing the excess was clearly needed, and so in later stages of this work we switched to the pressurized reservoir[9] both to prepare the sol-gel and to force it into and to remove the excess from the capillaries. In the context of one of the studies to

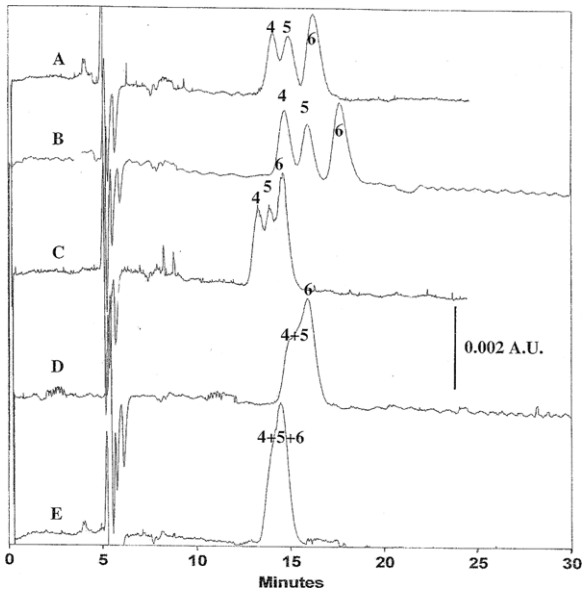

Figure 1. Separations from a study of the ratio of C8-TEOS—to—TEOS in the sol-gel mixture. Mobile phase: methanol-water (5:95, v/v), 25 mM acetate, 5 mM imidazole, pH 4.8. Sol-gel ratios: A = 0.1, B = 0.2, C = 0.4, D = 0.6, E = 0.8. The three sharp negative peaks are Mg(II), Zn(II), and Cd(II), respectively; those labeled 4, 5, and 6 are acetone, phenol, and p-nitrophenol, respectively.

determine the best mole ratio of C8-TEOS–to–TEOS spacer, we also looked at column to column reproducibility. Relative standard deviations of k for nitrophenol across all ratios from 0.1 to 0.8 in the series prepared were from 4–9%, with similar deviations for the resolution of nitrophenol from phenol, somewhat better than for manual sol-gel introduction and in general agreement with others who use the pressurized reservoir.[9] However, these data indicate that even with a more reproducible method of preparing the sol-gel and of introducing it into the capillaries, significant differences in chromatographic parameters from one column to the next persist.

A factor which may be one cause of poor reproducibility is the amount of unbonded precursor left in the porous capillary coating. It should be noted that if a newly prepared column was used with no methanol rinse, often the organic peaks were not seen at all, indicting too much stationary phase. But once sufficient methanol rinse was applied (several hundred μL) k values stabilized and peak sharpness improved, indicating that most nonbonded material was removed and coating morphology maximized.

As Li, Fries, and Malik point out in a recent review, the "design and synthesis of stationary phases with prescribed chromatographic and surface charge properties represent challenging tasks in contemporary CEC research."[8] It appears that significant effort is yet required to be able to reproducibly control this sol-gel technology and to uncover those synthetic parameters, which are critical to reproducible capillary coatings.

However, the main goal of this present effort was to demonstrate the ability to combine CZE and CEC to perform simultaneous separations of ions and neutral analytes both within and between classes, and so we present some examples of this work below. The ratio of precursors was 0.4 for the simultaneous separations described here.

Example Separations

Anion–organic separations generally used relatively soluble polar molecular analytes [acetone (unretained), phenol, nitrophenol]. Anions were chosen so that UV monitoring could be done at the same wavelength (250 nm) used for the organic species. A typical separation is shown in Figure 2. The mobile phase/run buffer contained a combination of the constituents normally used for CZE (pH 3 phosphate buffer) and that typically employed for reversed phase chromatography (10:90 (v/v) methanol-water). Note, that the ions come off the capillary before any of the organics, a typical situation, as they move more quickly than the EOF (indicated by the nonretained acetone analyte).

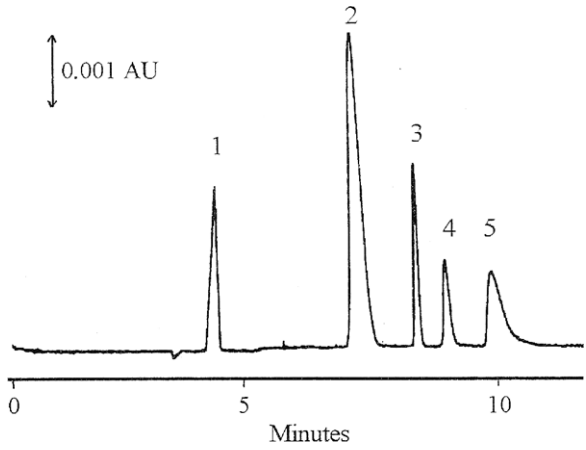

Figure 2. Separation of anions and neutral organic species on a 75 μM capillary with a stationary phase prepared from a sol-gel with a ODQAS—to—TEOS ratio of 0.4. Mobile phase: methanol-water (10:90, v/v), 25 mM phosphate, pH 3.0. Peaks: 1 = iodate, 2 = nitrite, 3 = acetone, 4 = phenol, 5 = p-nitrophenol.

As most metal ions do not absorb in the UV region needed for organic compounds, indirect detection was employed for cations.[14] Imidazole was used as the UV absorbing chromophore as its wavelength of maximum absorbance (214 nm, $\varepsilon = 5000$) is well below the typical 254 nm wavelength employed for organic species. In fact, ε for imidazole is zero at 254 nm. Again, a mixture of mobile phase/run buffer constituents was used (pH 4.8 acetate, 10:90 (v/v) methanol-water) with the imidazole chromophore. As was the case for the anion–organic mixtures, the cation grouping was well separated from the organics. This allowed a wavelength switching scheme to be used. Detection wavelength was set at 222 nm for the cations as this provided less baseline noise than at 214 nm. Once all the cations were past the detection window the wavelength was automatically switched to 254 nm. A typical separation is seen in Figure 3.

It was also of interest to attempt the simultaneous separation of metal cations and nonpolar organics. A similar mobile phase/run buffer was employed, 25 mM acetate and 5 mM imidazole. As nonpolar organic species are more highly retained than the polar organics by the wall coating, 70% (v/v) methanol-water was required. At this high methanol concentration the EOF could be as slow as 50 min, but with the addition of 1 mM sodium dodecylsulfate (SDS) the EOF is less than 20 min due to the presence of more "immobile" negative charges to augment the SiO^- sites.[15]

An example separation of several polynuclear aromatic hydrocarbons (PAHs) and three metal cations is shown in Figure 4. Again, the cations emerged from the capillary as a group, as did the PAHs. The

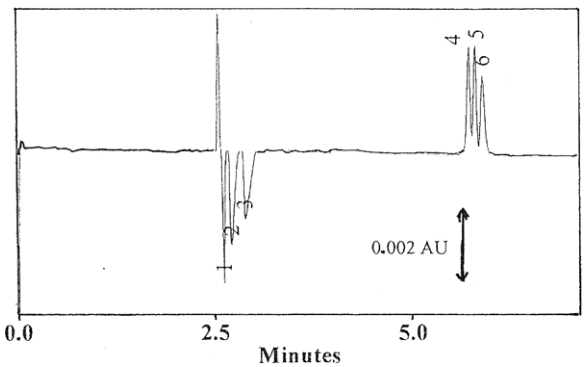

Figure 3. Separation of metals and polar organic species on a 75 μM capillary with a stationary phase prepared from a sol-gel with a C8-TEOS—to—TEOS ratio of 0.4. Mobile phase: methanol-water (10:80, v/v), 25 mM acetate, 5 mM imidazole, pH 4.8. Peaks: 1 = Mg(II), 2 = Zn(II), 3 = Cd(II), 4 = acetone, 5 = phenol, 6 = p-nitrophenol.

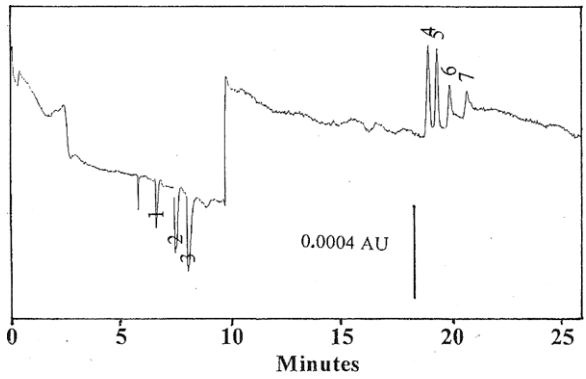

Figure 4. Separation of metal ions and PAHs on a 25 μM capillary with a stationary phase prepared from a sol-gel with a C8-TEOS—to—TEOS ration of 0.4. Mobil phase: methanol-water (70:30, v/v), 25 mM acetate, 5 mM imidazole, 1 mM SDS, pH 5.2. Peaks: 1 = Mg(II), 2 = Zn(II), 3 = Cd(II), 4 = acetone, 5 = naphthalene, 6 = phenanthrene, 7 = pyrene.

tailing seen for peaks 6 and 7 was often found for the larger nonpolar compounds on capillaries prepared in our lab, probably due to mass transfer effects in the interactions with the column coating.

The combination of CZE and CEC produced pH and mobile phase/run buffer effects that were not unexpected. For example, k values for nitrophenol were reduced four-fold, from about 0.5 to about 0.15, on increasing the methanol content of the mobile phase from 10% to 30% (v/v) on the ODQAS capillary, typical reversed phase behavior. Similar results were found with the C8-TEOS capillary, with k for nitrophenol decreasing about three-fold with a methanol increase from 10% to 30% (v/v). Similar behavior has been observed for ion–organic simultaneous separations on ion exchange HPLC columns, indicating again that separation mechanisms for ions and organics can operate independently of each other in mixed mode systems.[2–5]

The effect of methanol concentration on both metal ions and anions was what would be expected to result from an increase in viscosity of the run buffer as the methanol percentage increased.[16] For example, the electrophoretic mobility of Mg(II) on the C8-TEOS capillary decreased from about 26 cm^2/kVmin to about 18 cm^2/kVmin as the methanol concentration of the run buffer was increased from 10% to 30% (v/v). Similar behavior was exhibited by Zn(II) and Cd(II). The effect was less pronounced for anions on the ODQAS capillary, but still essentially what has been seen by others.[17] For example, the electrophoretic mobility of nitrite ion went from about 2.4 to about 0.2 cm^2/kVmin, as the methanol content of the run buffer (25 mM phosphate, pH 3.0) was changed

from 10% to 40% (v/v). Likewise, the mobility of iodate ion decreased from about 20 to about 14 cm^2/kVmin on going from 10% to 60% (v/v) methanol in the run buffer (25 mM phosphate, pH 3.0).

The effect of pH on retention of neutral organics was also examined on the ODQAS capillary. No significant change in k with pH was found for phenol with a pH change from 3 to 5. Changes in mobility for anions with pH were also investigated. As the pH was changed from 3 to 5 the

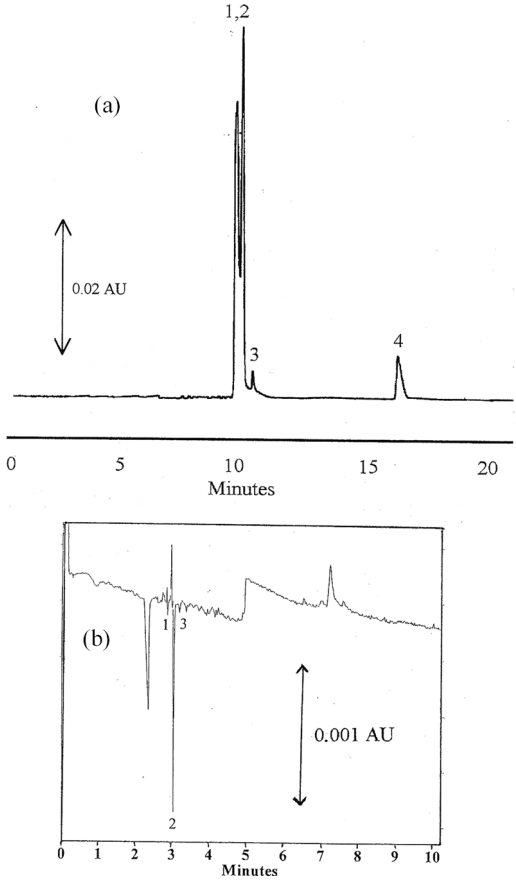

Figure 5. Separation of ions and neutrals of two commercial samples. (a) Water extract of a multivitamin, spiked with acetone, on a 75 µm ODQAC capillary. Mobile phase: methanol-water (10:90 v/v), pH 3.0. Peaks: 1 = vitamin C, 2 = acetone, 3 and 4 unknown. (b) A red wine (merlot) on a 75 µm C8-TEOS capillary. Mobile phase: methanol-water (10:90 v/v), 25 mM acetate, 5 mM imidazole, pH 4.8. Peaks: 1 = Ca(II) (possible), 2 = Mg(II), 3 = Zn(II). The large unnumbered negative peak may be Na^+. The organic (positive) peaks are not identified.

electrophoretic mobility of nitrite (pKa 3.2) increased from about 4 to about 25 cm^2/kVmin, as the nitrous acid is deprotonated causing increased mobility toward the anode. The mobility of iodate (pKa 0.77) also increased, changing from about 20 to about 39 cm^2/kVmin with a pH change from 3 to 5. This behavior is more difficult to understand as iodate should be deprotonated at both pH 3 and 5, and bears more investigation.

Column efficiency for organics in our hands was lower than reported by some others,[8,12] with N values in the range of 30,000 plates/m (nitrophenol on a 75 μm capillary) to 250,000 (phenol on a 25 μm capillary); and 20,000 (pyrene on a 75 μm capillary) to 230,000 (naphthalene on a 25 μm capillary). Efficiencies for anions on the ODQAS capillaries ranged from 10,000 to about 50,000 plates/m, while cations on the C8-TEOS capillaries ranged from 13,000 to 100,000 plates/m. These values are less then one would expect and may be due to some mass transfer effects on the wall coating. As expected, efficiencies were generally higher on the narrower bore capillaries.[12]

Example separations of a multivitamin tablet and a red wine are shown in Figure 5. In Figure 5(a), peak 1 is vitamin C, ascorbic acid, which is mostly protonated at pH 3 yet comes off the capillary before the unretained acetone due to its partial charge. The other peaks are not identified but are clearly interacting with the wall coating as their retention is beyond that of acetone. In Figure 5(B), peak 1 is Ca(II) and peak 2 is Mg(II). The organic peaks (positive) are not identified.

CONCLUSIONS

Despite the somewhat poor reproducibility and efficiency, wall coated capillaries offer promise for the simultaneous separation of ions and neutral organic species. Ions behave as they do in single mode capillary electrophoresis and organic neutrals as they do in reversed phase HPLC. Examples cited show that using wall coated capillaries and a combined run buffer/mobile phase allows for separations using both CZE and CEC simultaneously. If the column preparation techniques can be improved to enhance column to column reproducibility and efficiency, then simultaneous ion-neutral separations driven electrophoretically may present advantages over HPLC.

REFERENCES

1. Muenter, M.M.; Stokes, K.C.; Obie, R.T.; Jezorek, J.R. Simultaneous separation of inorganic ions and neutral organics on ion-exchange stationary phases. J. Chromatogr. A. **1999**, *844*, 39–51.

2. Dean, T.H.; Jezorek, J.R. Demonstration of simultaneous anion exchange and reversed phase behavior on a strong anion exchange column. J. Chromatogr. A. **2004**, *1028*, 239–245.
3. Gu, R.-F.; Jezorek, J.R. Demonstration of simultaneous cation-exchange and reversed-phase mechanisms on a strong cation-exchange column. J. Chromatogr. A. **2001**, *919*, 21–28.
4. Jezorek, J.R.; Wagner, K.A. Simultaneous separation of metal ions and neutral organics by reversed-phase ion-pair liquid chromatography. J. Chromatogr. A. **1997**, *776*, 221–231.
5. Maness, S.J.; Jezorek, J.R. Simultaneous separation of neutral organics and inorganic anions by coupling reversed-phase and ion-interaction chromatography. J. Liq. Chromatogr. & Rel. Technol. **2005**, *28*, 2921–2930.
6. Terabe, S. Electrokinetic chromatography: an interface between electrophoresis and chromatography. Trends Anal. Chem. **1989**, *8*, 129–134.
7. Colon, L.A.; Guo, Y.; Fermier, A.M. Capillary electrochromatography. Anal. Chem. **1997**, *69*, 461A–467A.
8. Li, W.; Fries, D.P.; Malik, A. Sol-gel stationary phases for capillary electrochromatography. J. Chromatogr. A. **2004**, *1004*, 23–52.
9. Rodriguez, S.A.; Colon, L.A. Investigations of a sol-gel derived stationary phase for open tubular capillary electrochromatography. **1999**, *397*, 207–215.
10. Hayes, J.D.; Malik, A. Sol-gel open tubular ODS columns with reversed electroosmotic flow for capillary electrochromatography. Anal. Chem. **2001**, *73*, 987–996.
11. Constanin, S.; Freitag, R. Preparation of stationary phases for open tubular capillary electrochromatography using the sol-gel method. J. Chromatogr. A. **2000**, *887*, 253–263.
12. Guo, Y.; Colon, L.A. A stationary phase for open tubular liquid chromatography and electrochromatography using sol-gel technology. Anal. Chem. **1995**, *67*, 2511–2516.
13. Rodriguez, S.A.; Colon, L.A. Study of the solution in the synthesis of a sol-gel composite used as a chromatographic phase. Chem. Mater. **1999**, *11*, 754–762.
14. Hjerten, S.; Elenbring, K.; Kilar, F.; Liao, J.-L.; Chen, A.J.C.; Siebert, C.J.; Zhu, M.-D. Carrier-free zone electrophoresis, displacement electrophoresis and isoelectric focusing in high performance electrophoresis apparatus. J. Chromatogr. **1987**, *403*, 47–61.
15. Chen, M.; Cassidy, R.M. Bonded-phase capillaries and the separation of inorganic ions by high-voltage capillary electrophoresis. J. Chromatogr. **1992**, *602*, 227–234.
16. Shi, Y.; Fritz, J.S. New electrolyte system for the determination of metal cations by capillary zone electrophoresis. J. Chromatogr. **1994**, *671*, 429–435.
17. Kaniansky, D.; Masar, M.; Marak, J.; Bodor, R. Capillary electrophoresis of inorganic anions. J. Chromatogr. **1999**, *834*, 133–178.

Received April 3, 2008
Accepted June 11, 2008
Manuscript 6337